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COMBINING ABILITY OF ALFALFA CLONES DIFFERING IN TOLERANCE TO FREQUENT HARVEST

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ABSTRACT

Frequent harvest-tolerant (T) and -sensitive (S) clones were selected from spaced plants of eight local and exotic non-dormant alfalfa varieties that survived an extended frequent harvest regime. Clones were established in isolation for the production of open-pollination (OP) seed for testing their general combining ability (GCA). OP seeds of eighty (T) clones, bulks of (S) clones from eight varieties and the parent varieties were sown in Jan 2000 in separate field trials for evaluating performance over six monthly harvests (frequent) and four 45-day harvests (infrequent) during the period August 15, 2000 to Jan 15, 2001. Progenies of T and S clones were highly variable but were similar in average response to harvesting schedule and showed less reduction in yield from frequent harvest than their parent varieties. Significant variation was detected between progenies in each group under infrequent harvest and between T progenies alone under frequent harvests. Significant differences in GCA between clones were observed for dry matter yield and related traits in each harvesting schedule with a significant clone x schedule interaction and no correlation of total yield of progeny between harvesting schedules. The per se performance of clones in spaced nursery was also not correlated with OP progeny performance. An 8-clone synthetic selected for GCA under frequent harvest was predicted to exhibit an 8% yield reduction from infrequent to frequent harvest, and 17% less yield under infrequent harvest than a similar synthetic selected on the basis of GCA under infrequent harvest. Heritability estimates of total dry forage yield were generally slightly higher under infrequent than frequent harvest. The progress of frequent harvesting was associated with increased heritability of crown bud score in S clones and plant height and vigor score in T clones, suggesting differences among clonal groups in means of coping with harvest stress.

Key words: *Non-dormant alfalfa, Harvest schedules, Frequent harvest tolerance, Combining ability, Progeny test, Heritability.*

Abbreviation: *T= Frequent harvest-tolerant, S= Frequent harvest sensitive, OP =Open- pollination, GCA =General combining ability.*

INTRODUCTION

Frequent harvesting of immature non-dormant alfalfa produces high-quality forage but reduces long-term forage production and accelerates stand decline (Rammah and Hamza 1980 and Marble, 1989) Improved alfalfa cultivars adapted to frequent harvests are not currently available. Early studies (Chatterton *et al* 1977 and Veronesi *et al* 1986) pointed out the difficultly of developing cultivars that yield comparably under frequent and less frequent harvests. Despite the presence of significant variability for yield and tolerance to frequent harvests among alfalfa clones, no clones were identified which could yield as well under frequent as under infrequent harvests (Chatterton *et al* 1977). Recurrent phenotypic selection for tolerance to frequent harvests was effective in improving persistence and yield under frequent harvests, but failed to overcome the decrease in dry matter yield caused by frequent harvests (Veronesi *et al* 1986). However, there are indications that some non-dormant local ecotypes of alfalfa in the Near East may be adapted to frequent harvests (Marble, 1989 and Al-Doss, 2001). In an earlier study (Radwan *et al* 2003), we identified frequent-harvest tolerant (T) and sensitive (S) clones from spaced plants of local and introduced cultivars of non-dormant alfalfa. Forage yield of spaced plants of some T clones was not significantly reduced from infrequent to frequent harvests during a period of 120 d of harvest management. Such clones would be useful for developing synthetic varieties adapted to variable harvest frequency, if they possess high general combining ability (GCA) regardless of harvesting system.

The objectives of the present study were to: i) estimate GCA of forage yield of T and S clones under contrasting harvesting schedules, ii) correlate performance of clones from spaced-plants with progeny performance in dense stands under frequent and infrequent harvests and iii) predict the yield of synthetics selected under each harvesting system.

MATERIALS AND METHODS

Vigorus and weak plants in a spaced nursery that survived an extended regime of frequent harvests were selected from eight alfalfa varieties including Midia, Marina, ORO and Medina from France, WL 605 from USA, and Ismaelia 1, Ismaelia 94 and Siwa 1 from Egypt. The plants were transferred to the greenhouse and vegetatively propagated. Clones of selected vigorus plants were designated as tolerant (T) and those of weak plants as sensitive (S) to frequent harvests.

In April 1998, established propagules from each clone were transplanted in the field at Giza Research Station (ARC) at 25 cm spacing in single-row plots, 2 m long, arranged in a simple lattice design with two replications. Only 145 (T) and 114 (S) clones were successfully established in the field. Established plants flowered in June, 1998 and were allowed to produce seed by open-pollination for the progeny test. Sufficient seed was harvested in October 1998 from 80 tolerant clones. Sensitive clones produced little seed, therefore seed from all sensitive clones of each variety were bulked together to represent the sensitive clones.

OP seed of T clones, seed bulks of S clones and parent varieties were sown at the Experimental Station of the Faculty of Agric., Cairo Univ. in January 2000 in two adjacent trials, one for testing performance under a schedule of frequent harvests (six monthly harvests) and the other for an infrequent harvests (four 45-day harvests) schedule. Trials were arranged as RCB with two replications. Plots consisted of single rows 3m long, 50 cm wide sown at the rate of 33 Kg/ha. On July 15, trials were uniformly harvested then both harvesting schedules began August 15, 2000 and ended in Jan 15, 2001. At harvest, data were collected on plant height, fresh forage yield and number of crown buds and regrowth vigor one week after harvest, both scored on a 1-5 scale, (1=lowest). Dry forage yield was estimated from the percentage of dry matter obtained by drying forage samples from each plot. Data of each trial were subjected to regular analysis of variance and a combined analysis of variance of both trials was computed for total dry yield of all harvests. GCA effects of clones for total yield under each harvesting schedule were estimated as the deviation of the progeny mean of a clone from the general mean of all progenies. Estimates of heritability (h^2) for traits under each harvesting schedule were computed from components of variance derived from ANOVA for each harvest as follows: $h^2 = \sigma^2_t / (\sigma^2_t + \sigma^2_e/r)$ for tolerant progenies and $h^2 = \sigma^2_s / (\sigma^2_s + r\sigma^2_e/r)$ for sensitive progenies, where σ^2_t, σ^2_s and σ^2_e are genotypic variances for tolerant and sensitive clones and error, respectively.

RESULTS

Variation between OP progenies.

Analysis of variance for each trial indicated that progenies of T and S clones were often significantly taller, had more crown buds and greater regrowth vigor than their parents. Results of the combined analysis of variance of total dry forage yield under frequent and infrequent harvests are shown in Table (1). Harvesting schedule significantly affected dry yield (F= 155.6**) of tested genotypes. Average dry yield over genotypes exhibited a significant 18.3% reduction from infrequent to frequent harvesting.

Table 1. Mean squares from combined ANOVA of total dry forage yield of open-pollinated progenies of alfalfa clones and parent varieties, under two harvesting schedules, 2000-2001.

S.O.V.	d.f	Total dry yield
Schedules (Sc)	1	933.3**
Genotypes (G)	95	18.8**
Parents (P)	7	11.6
Progenies, T clones	79	18.2*
Progenies, S clones	7	19.4
T vs. S	1	3.7
P vs. (T + S)	1	113.3
G x Sc.	95	11.0**
P x Sc.	7	12.0
T x Sc.	79	10.7**
S x Sc.	7	15.0*
Residual	2	8.3
Error	190	6.0

*, ** significant at 5 and 1% level, respectively.

Average dry yields of T and S progenies and parents were reduced by 17.0%, 15.1% and 25.4%, respectively, from infrequent to frequent harvesting (Table 2). Progenies of T clones derived from local varieties yielded almost equally under both systems of harvesting, whereas those from exotic varieties exhibited a significant reduction in yield under frequent harvestin (Table 2). Wide variation in the effect of the harvesting schedule on yield performance was also noted between progenies within the T and S clonal groups according to originating variety, and between the parent varieties themselves.

However, despite variation in response to harvesting schedule, average dry yields of S progenies were not significantly affected by harvest frequency except for progenies derived from the Midia variety which exhibited exceptionally high performance under infrequent harvest (Table 2)

The parent varieties were not significantly different in yield, showed less variation in response to frequent harvesting, but differed widely in response to infrequent harvests (Table 2). The greatest significant yield reduction from infrequent to frequent harvest was shown for varieties ORO (40.2%), Midia (32.9%), WL 605 (32.4%) and Medina (31.0%), whereas progenies of T clones from the same varieties exhibited dry yield reduction of 18.6, 31.8, 26.8 and 30% in the same order, suggesting a degree of association between clones and originating variety in response to frequent harvests (Table 2). Dry yield of the three local varieties and progenies of

Table 2. Total dry forage yield t ha^{-1} of OP progenies of tolerant and sensitive alfalfa clones and their parent varieties under frequent and infrequent harvests, 2000-2001.

	Var.	No. clones	Freq.	Infr.	% reduction @
Tolerant clones progeny	Midia	12	13.8	20.2**	-31.8**
	Marina	12	13.6	15.9*	-14.9*
	ORO	12	16.7	20.5**	-18.6**
	Medina	8	11.7	16.7**	-30.0**
	WL 605	12	12.4	16.9**	-26.8**
	Ismaelia 1	11	16.7	17.1	-2.8
	Ismaelia 94	6	14.8	14.8	0.0
	Siwa 1	7	13.6	13.3	1.8
Weighted mean		80	14.2a	17.3a**	-17.0**
Sensitive Clones progeny	Midia	1	14.0	25.0*	-43.8*
	Marina	1	19.8	16.2	22.1
	ORO	1	16.2	15.9	1.5
	Medina	1	15.2	19.3	-21.0
	WL 605	1	15.2	18.1	-15.8
	Ismaelia 1	1	15.2	14.0	8.5
	Ismaelia 94	1	10.7	14.5	-26.3
	Siwa 1	1	12.1	14.0	-13.6
Mean		8	14.8a	17.4a	-15.1
Parents varieties	Midia		12.6	18.8	-32.9*
	Marina		13.6	15.7	-13.6
	ORO		12.4	20.7	-40.2*
	Medina		9.5	13.8	-31.0
	WL 605		10.9	16.2	-32.4*
	Ismaelia 1		12.6	15.2	-17.2
	Ismaelia 94		11.9	11.4	4.2
	Siwa 1		12.4	15.5	-20.0
Mean		8	11.9b	15.9a	-25.4
General mean			13.6	16.8**	-19.2**

*, ** Significant at 5% and 1%, respectively..

@ Yield difference between frequent and infrequent as % of infrequent harvests.

Group means in one column having the same letter are not significant different at P=0.05

their T clones were not significantly affected by the frequent harvests, at least within the duration of the study. However, these varieties were generally less productive under infrequent harvest compared with some of the exotic cultivars (e.g. ORO).

General combining ability

Progenies of T clones exhibited a wider range of variation in dry matter yield than S clones under frequent harvests (19.6 to 8.4 vs. 19.7 to 10.8 t ha^{-1}), and infrequent harvests (24.3 to 10.9 vs. 25.1 to 14.0 t ha^{-1}). Eleven T clones showed significant GCA effect for total dry matter yield under infrequent, compared to only seven clones under frequent harvests.

However, only two clones were common to the set of clones identified in each system indicating that T clones high in GCA under frequent harvests were generally of lower GCA under infrequent harvests and *vice verse* (Table 3). For S composites, significant GCA effect was only detected for the Marina variety under frequent harvest and for Ismaelia 1 variety under infrequent harvest.

Table 3. GCA effects of T and S alfalfa clones with the highest yielding OP progenies under frequent and infrequent harvests, and corresponding yields in opposite schedule, 2000-2001.

Frequent				Infrequent			
Var./clone	GCA effect $t\ ha^{-1}$	Dry yield $T\ ha^{-1}$		Var./clone	GCA Effect $t\ ha^{-1}$	Dry yield $t\ ha^{-1}$	
		Freq.	Infr.			Infr.	Freq.
Ismaelia 1/ 8	5.3*	19.6	16.7	Midia / 4	7.0*	24.3	15.5
ORO / 2	4.5*	18.7	19.8	ORO / 9	6.1*	23.5	18.7
ORO / 9	4.4*	18.7	23.5	Midia / 3	5.7*	23.1	11.8
Ismaelia 1 / 5	4.2*	18.5	18.2	ORO / 19	5.8*	23.1	15.9
ORO / 7	3.5*	17.8	21.8	Midia / 17	5.6*	22.8	15.3
ORO / 5	3.3*	17.5	19.7	Midia / 7	5.4*	22.7	13.3
Ismaelia 94 / 15	3.1*	17.5	17.3	Midia / 15	4.7*	22.0	13.9
Marina [a]	5.5*	19.7	18.1	Ismaelia 1 [a]	7.7*	25.1	15.2
Mean		17.9	19.4	Mean		23.3	15.0**

* Significant difference from general mean of progenies.
[a] S composite.

The spearman's correlation coefficient between yields of progenies (88 progenies) under the two harvesting systems was very low and insignificant (r=0.11). Though the correlation of ranks of progenies under both schedules was rather strong and significant (r=0.70*, T and 0.79*, S), the highest yielding eight clonal progenies were ranked quite differently in each harvest schedule, with only one clone (ORO 9) common to both harvest treatments (Table 3).

Performance of spaced clones vs. drilled progeny

The *per se* yield of (44) T and S clones in spaced nursery (Radwan *et al* 2003) was compared with the performance of their OP progenies in drilled seeding. A very weak correlation was detected between clones and progenies (r= 0.001 for frequent and 0.12 for infrequent harvests), suggesting that selection in spaced plants is not effective in identifying clones with high GCA.

However, data (not presented) comparing the *per se* performance of the highest yielding clones and their progenies generally suggest that selection of T clones from frequently harvested spaced plants identifies clones whose OP progenies yield well under both frequent and infrequent harvests, but progenies of clones selected under infrequent harvests yield well only under infrequent harvest. In contrast, selection of the best S clones under frequent harvests identifies clones whose progenies yield more under infrequent than frequent harvests.

Heritability

Under frequent harvest, heritability of dry yield of both T and S clones was negative at the first harvest and increased with harvest frequency indicating that intra-group genetic variability was limited at the onset of the harvesting schedule and became more expressed as harvesting stress increased. For total dry yield, similar heritabilities were obtained for T and S clones under both harvesting schedules but estimates were generally slightly higher under infrequent than frequent harvests (Table 4).

Table 4. Estimates of heritability for traits of two harvests and total dry matter yield of tolerant and sensitive progenies under two harvest schedules, 2000-2001.

Traits	Tolerant progenies				Sensitive progenies			
	Freq.		Infreq.		Freq.		Infreq.	
	1st cut	Last cut	1st cut	Last cut	1st cut	Last cut	1st cut	Last cut
Dry yield	0.00	0.74	0.63	0.71	0.00	0.62	0.67	0.81
Plant height	0.00	0.64	0.62	0.26	0.35	0.00	0.57	0.00
Crown buds score	0.44	0.40	0.48	0.52	0.34	0.86	0.70	0.33
Regrowth vigor score	0.00	0.68	0.62	0.61	0.29	0.29	0.61	0.43
Total dry yield	0.52		0.62		0.63		0.72	

The progress of frequent harvests was associated with increased heritability of crown bud score of S progenies, and plant height and regrowth vigor score of T progress (Table 4). A reverse trend was shown for heritability estimates of the same traits under infrequent harvests (Table 4). Thus the expression of intra-group genetic differences was enhanced or reduced for different yield-related traits in each group according to the level of harvest stress. This might indicate differences between clonal groups in strategies of coping with harvest stress.

DISCUSSION

T and S clones respectively represent vigorus and weak plants that survived through frequent harvests. Under spaced-planting, the T clones tolerated frequent harvest better than S clones but both groups were equally productive under normal harvesting (Radwan *et. al* 2003). However, in the present study, OP progenies of T and S clones were not significantly different in average performance, and their yield showed similar decline with successive harvests. However, both groups were significantly higher yielding than their parent varieties, indicating that plant selection either for survival or vigor under frequent harvesting might enhance potential productivity (Veronesl *et al* 1986, Purves and Wynn-Williams 1994).

Total dry matter yield of three exotic varieties and progenies of T clones originating from all exotic varieties were significantly reduced from infrequent to frequent harvests, whereas yield was not significantly reduced by the frequent harvest for the local varieties and their T progenies. In contrast, dry yield of progenies of all S clonal composites except those of cv. Medina was not significantly decreased by frequent harvest. This contrasting behavior of T and S progenies might suggest that their parent clones represent different physiologic and genetic norms. However, the weak and insignificant correlation between the *per se* performance of clones and their sexual progenies might suggest that differentiating clones to T vs. S on the basis of vigor of surviving plants is not a suitable procedure for judging tolerance to frequent harvesting. Wilsie and Skory (1948), also reported insignificant correlation between the yields of clones and their OP progenies.

An interesting finding of the present work is the significant GCA harvest schedules interaction, resulting in very few clones with high GCA under both harvesting schedules, or clones of higher GCA under frequent than infrequent harvest. Though these two types of clones were of very low frequency, there appears a greater potential for identifying them within populations of the local varieties and the introduced Marina variety.

For traits determined primarily by additive genetic effects, only one synthetic variety can be produced comprising parents with good GCA (Hill *et al.*, 1972). Since the performance of this synthetic may be predicted from the average performance of all crosses among its component clones (Peadrson and Elling 1961), performance may well be predicted, though less accurately, from the average performance of the OP progenies of its parent clones. On this basis, a synthetic developed from 8 clones with the highest GCA under frequent harvest was predicted to yield 17.9 t ha^{-1} under frequent and 19.4 t ha^{-1} under infrequent harvest (a reduction in yield of 7.7%). But combining 8 clones of the highest GCA under infrequent harvest would produce a synthetic with a predicted yield of 23.3 t ha^{-1} under infrequent and 15.0 t ha^{-1} under frequent harvest (a reduction of 35.6%).

These predictions suggest that direct selection under frequent harvest would improve yield of synthetics under frequent harvest more than indirect selection, but would sacrifice some (17.0%, 17.9 vs. 19.4 t ha^{-1}) of the yield potential from selection under normal harvest, which is in general agreement with Veronesi *et al* (1986).

Frequent harvesting schedules represent a stressful management expected to exert different effects on different plant genotypes according to the pattern of storage of root and crown reserves and the stage at which harvest is made. In addition, a frequent harvesting regime could be applied only at seasons where alfalfa growth rate is high, while a normal harvesting regime that recovers the plants vigor is applied during the seasons of slower growth. In this case genotypes would be sought that grow rapidly and utilize a minimum of root and crown reserves to cope with frequent harvests, and are capable of storing adequate organic reserves during seasons of slow growth to enhance plant survival. Non-dormant alfalfa varieties show differences in the pattern of growth across seasons (Rammah and Hamza 1980). However, there is currently a paucity of information correlating growth of non-dormant clones with variation in root and crown reserves, especially in light of accumulating evidence of the less important role of TNC reserves in regrowth of non-dormant alfalfa (Al-Doss 2001).

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القدرة على التآلف لسلالات البرسيم الحجازي المختلفة في تحملها للحش المتكرر

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تم انتخاب عدة سلالات متحملة للحش المتكرر وأخرى حساسة للحش المتكرر وذلك من ثماني أصناف محلية ومستوردة من البرسيم الحجازي . زرعت هذه السلالات في مكان معزول للتلقيح فيما بينها لانتاج البذرة مفتوحة التلقيح. تم زراعة البذرة مفتوحة التلقيح لثمانية سلالات متحملة للحش المتكرر ، مخلوط البذرة مفتوحة التلقيح للسلالات الحساسة للحش من كل صنف الإضافة إلى الأصناف الأبوية وذلك فى تجربة لتقييم السلوك خلال ستة حشات شهرية (حش متكرر) وأربعة حشات كل ٤٥ يوم (حش غير متكرر)

وجد أن أنسال السلالات المتحملة والحساسة للحش اختلفت فيما بينها فى استجابتها لميعاد الحش وقد أظهرت أقل انخفاض فى المحصول عند الحش المتكرر وذلك بمقارنتها مع الأصناف الأبوية. لوحظ أن الأنسال فى كل مجموعة قد تباينت معنويا تحت نظام الحش العادى (غير المتكرر) وكذلك اختلفت أنسال السلالات المتحملة للحش المتكرر فقط تحت نظام الحش المتكرر. كانت الاختلافات فى القدرة العامة للتآلف معنوية بين السلالات وذلك بالنسبة للمحصول الجاف والصفات المتعلقة به فى كلا ميعادي الحش، كما كان تفاعل السلالات مع ميعاد الحش معنوى . أعطت السلالات المنتخبة للقدرة العامة على التآلف تحت نظام الحش المتكرر أعطت نسلا تركيبى يتوقع أن يظهر ٨ % نقصا فى المحصول فى الحش المتكرر عنه فى الحش غير المتكرر (العادى) ، ١٧ % انخفاض فى المحصول تحت الحش العادى وذلك عند المقارنة مع السلالات المنتخبة للقدرة على التآلف تحت نظام الحش العادى (غير المتكرر) . كانت تقديرات المكافئ الوراثى للمحصول الجاف الكلى بصفة عامة عالية نسبيا تحت نظام الحش العادى عنها تحت نظام الحش المتكرر . ارتبطت الزيادة فى الحش المتكرر مع زيادة المكافئ الوراثى بالنسبة لصفات معدل براعم التاج فى السلالات الحساسة للحش ، ارتفاع النبات و قوة النمو فى السلالات المتحملة للحش .

مجلد المؤتمر الرابع لتربية النبات–الإسماعيلية ٥ مارس ٢٠٠٥

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